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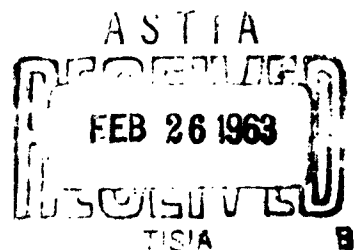
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PAROTID FLUID AND SERUM GLUCOSE LEVELS  
AND PAROTID FLUID FLOW RATE AS RELATED  
TO DENTAL CARIES STATUS

TECHNICAL DOCUMENTARY REPORT NO. SAM-TDR-62-140

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USAF School of Aerospace Medicine  
Aerospace Medical Division (AFSC)  
Brooks Air Force Base, Texas



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## **FOREWORD**

This report was prepared by the following personnel at the USAF School of Aerospace Medicine\* and Tufts University—School of Dental Medicine†:


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## ABSTRACT

Parotid fluid samples were collected without exogenous stimulation from 507 systemically healthy young adult males. Blood was drawn from each participant and subjects were classified as to dental caries experience (DMFS). Parotid fluid flow rate was recorded, and serum and parotid fluid were analyzed for glucose concentration by the glucose oxidase procedure. The presence of glucose in parotid fluid was confirmed. Neither parotid fluid flow rate, parotid fluid glucose level, nor serum glucose concentration was found to be related to caries experience. For all participants, the mean for each of the three variables was 0.042 (S.D.  $\pm$  0.036) ml./min., 0.75 (S.D.  $\pm$  0.68) mg. %, and 84.8 (S.D.  $\pm$  7.76) mg. %, respectively.

This technical documentary report has been reviewed and is approved.

  
ROBERT B. PAYNE  
Colonel, USAF, MSC  
Chief, Operations Division

## PAROTID FLUID AND SERUM GLUCOSE LEVELS AND PAROTID FLUID FLOW RATE AS RELATED TO DENTAL CARIES STATUS

### 1. INTRODUCTION

The importance of intraoral carbohydrates in the caries process has been one of the most frequently discussed subjects in the dental literature. Bunting and his group (1-4) reported many years ago that the institution of a low-sugar diet arrested caries in the majority of cases. These studies led to the conclusion that diet controlled caries by altering the environment of teeth rather than by making them more resistant to caries (2). Becks (5, 6) initiated carbohydrate restriction in 1,250 subjects with rampant caries and found that, in about two-thirds of the cases, no new lesions developed in the year during which the restricted diet was followed. While there is convincing evidence that the production of acid from carbohydrate is not the entire explanation of the carious process, the majority opinion is that the primary lesion is produced by acid decalcification of the inorganic portion of the enamel and that the degradation of carbohydrates to lactic acid is of primary importance in this regard.

The development of an enzymatic laboratory method with a specificity for glucose has made it possible to establish that this substance is actually present in parotid fluid in measurable amounts (7, 8). It was of interest to determine whether or not levels of this biologically important endogenous carbohydrate in body fluids were related to dental caries status.

### 2. MATERIALS AND METHODS

Subjects were 507 males between the ages of 17 and 22 years. Each had been proved physically fit for military service by recent medical examination. Environmental condi-

tions and dietary and emotional exposure were similar for all subjects. Oral health status was not a factor in the selection of subjects.

Collection of parotid fluid was initiated between 7:30 and 8:00 a.m., this being 2 to 2½ hours after breakfast. A parotid cap (9) was placed over the orifice of the duct of the right gland with an absolute minimum of manipulation, and parotid fluid was collected over a period of at least 2 hours. No exogenous stimulants of any type were employed; on the contrary, every precaution was taken to minimize stimulation as much as possible. During the sampling period the patients were seated comfortably in a semi-isolated section of an air-conditioned laboratory that was kept free from extraneous disturbances. Subjects were allowed to write letters and to peruse selected reading material in an effort to minimize psychic glandular stimulation. All participants were carefully supervised to prevent talking and unnecessary movement and to assure that they remain awake and alert. Parotid fluid was collected in graduated tubes and volume was read to the nearest 0.05 ml. A venous blood sample was collected from each participant between 10:00 and 10:30 a.m. and, as soon as a clot was formed, the serum was separated and retained. The glucose concentration of both fluids was determined enzymatically with glucose oxidase by methods based on the contributions of Keston (10) and of Saifer and Gerstenfeld (11). Serum, 0.2 ml., was taken to 4.0 ml. with water and the proteins were precipitated by the addition of 2.0 ml. each of 2% zinc sulfate and 0.12 normal NaOH. This mixture was filtered and 1.0 ml. of filtrate was exposed to 4.0 ml. of Glucostat<sup>1</sup> reagent at

<sup>1</sup>Worthington Biochemical Corp., Freehold, N. J.

37° C. for one hour. The reaction was halted with acid, and the developed color was read at 400 m $\mu$  in a Beckman DU spectrophotometer. In the parotid fluid determination, 0.5 ml. of the sample was added to 0.5 ml. of 2% zinc sulfate, and 0.5 ml. of 0.12 normal NaOH was added to this mixture. Filtrate and Glucostat were mixed in equal portions in the saliva procedure and color was read in microcuvettes. The performance of each method was evaluated as to reproducibility and recovery ability.

Subjects were divided into six dental status groups based upon the sum of decayed, missing, and filled tooth surfaces (DMFS) noted. Thorough clinical examinations were supplemented by bitewing roentgenograms. Third molars were excluded in all instances. Each subject possessed at least 20 teeth and none presented evidence of advanced periodontal disease.

### 3. RESULTS

The ability of the enzyme procedure to recover glucose added to serum is outlined in table I. The mean recovery rate was 103%. Duplicate glucose determinations were carried out on 20 sera (mean glucose, 82.3 mg.%) to test method reproducibility. The average difference between duplicates was 0.96 mg. %, the standard deviation of this difference was 0.81 mg.%, and the coefficient of variation for the method was 0.98%.

When 20 parotid fluid samples were analyzed in duplicate for glucose content a

mean of 0.80 mg.% was found. The average difference between duplicates was 0.04 mg.%, and the standard deviation of this difference was 0.033 mg.%. The coefficient of variation for the salivary procedure was 4.1%. The method performed well in recovering added glucose. Glucose was added to portions of a parotid fluid pool that contained 0.77 mg./100 ml. To 0.3773, 0.3465, and 0.3080 mg. of glucose from the pool, 0.20, 1.00, and 2.00 mg. of glucose, respectively, were added from a 20 mg./100 ml. stock solution. Glucose in the amounts of 0.570, 1.345, and 2.305 mg. were found in these three solutions with a resultant mean added glucose recovery rate of 99.7%.

Figure 1 demonstrates that parotid fluid flow rate was independent of DMFS status. This confirms our past observations under similar experimental conditions (12, 13). The mean of 0.036 ml./min. found for the 31 to 40 DMFS group differed from the means for the 11 to 20 and 41 to 50 groups but, since these differences were significant only at the .05 level and since no pattern of differences was evident, they were held due to chance. The mean for all participants was 0.042 (S.D.  $\pm$  0.036) ml./min.

For serum glucose, there were no significant differences between DMFS groups (fig. 2). The serum glucose mean for the 507 subjects was 84.8 (S.D.  $\pm$  7.76) mg.%. For parotid fluid glucose, the mean of 0.57 mg.% for the 41 to 50 DMFS group was significantly lower than the means for the 0 to 10, 11 to 20, and 21 to 30 DMFS groups (fig. 3). As with the

TABLE I  
*Recovery of added glucose (mg.)*

Flask No.	(a) From serum pool	(b) Added from stock solution	(c) Found	(d) Recovered (c-a)	(e) Difference (d-b)	Percent recovery (d/b $\times$ 100)
1	41.250	—	41.25	—	—	—
2	40.425	10.00	51.10	10.675	+ 0.675	107
3	38.775	30.00	70.00	31.225	+ 1.225	104
4	37.125	50.00	88.35	51.225	+ 1.225	102
5	35.475	70.00	105.55	70.075	+ 0.075	100



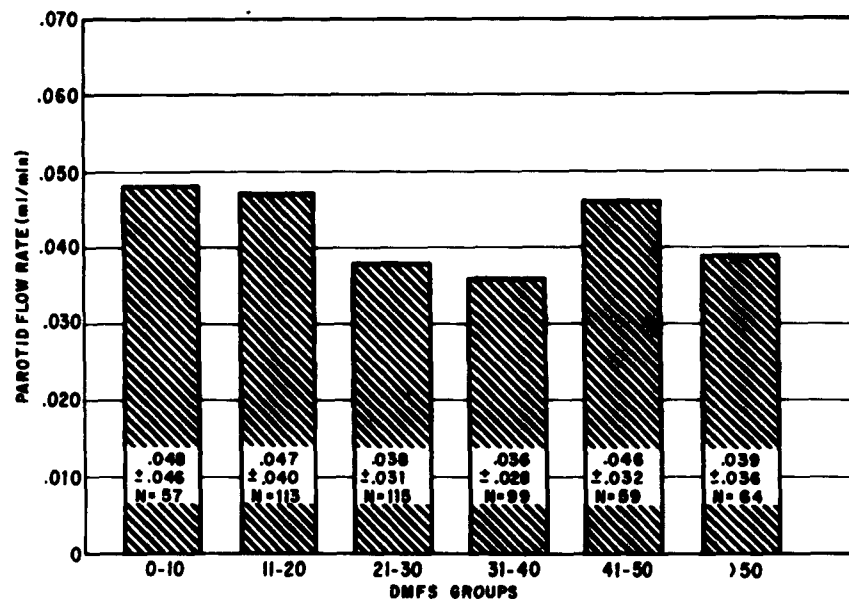


FIGURE 1

*Means ± S.D. for parotid flow for DMFS groups.*

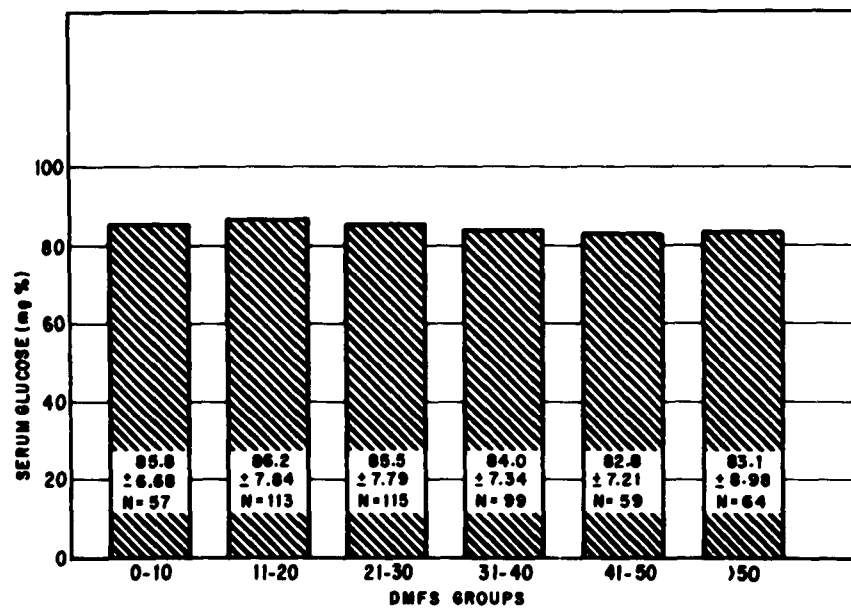


FIGURE 2

*Means ± S.D. for serum glucose for DMFS groups.*

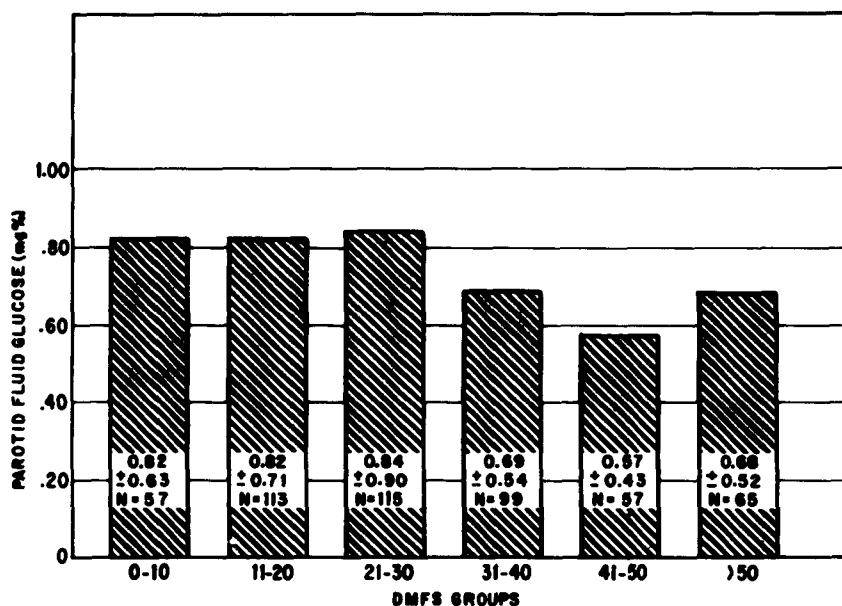


FIGURE 3

*Means  $\pm$  S.D. for parotid fluid glucose for DMFS groups.*

flow rate results, no pattern was discernible, and this observation was held probably due to chance. The overall mean for all subjects was 0.75 (S.D.  $\pm$  0.68) mg.%.

#### 4. DISCUSSION

Many investigations have been conducted to ascertain the presence or absence of glucose in saliva. The investigators who have claimed its presence have attempted to determine the mechanisms responsible for the control of the salivary sugar level primarily by altering the blood level and following the response in saliva. The extensive disagreement in past results has been due primarily to the lack of an analytic method with a specificity for glucose. In 1960, Shannon and Prigmore (14) evaluated the performance of an automatic method of parotid

fluid glucose determination and reported a glucose mean of 11.14 mg.%. In the same year, Shannon et al. (7) found that this automated ferricyanide method gave falsely high results when compared to the enzymatic procedure which takes advantage of the exquisite specificity of glucose oxidase. These authors concluded that there was a measurable level of glucose in human parotid fluid and that the level was sensitive to the intravenous administration of glucose. In 1962, Englander et al. (8) reported measurements of parotid fluid glucose by the glucose oxidase procedure in both normal and diabetic subjects. Wolcott and Weber (15) studied parotid fluid by the anthrone procedure in an attempt to develop a glucose tolerance procedure. Weber et al. (16) found three times as much carbohydrate in the parotid fluid of caries-rampant in-

dividuals as in caries-immunes and implied that this finding also held true for glucose by stating that glucose oxidase / anthrone ratios compared favorably. No glucose data were presented.

The present study confirms the presence of glucose in parotid fluid. Although the mean for this variable was only about 1% of the serum glucose mean, there was a measurable level in the parotid fluid of each participant. It seems quite clear that the endogenous

salivary glucose is not a fermentable carbohydrate of importance in the decay process. The concentration of glucose is so low that its presence is of little importance when compared to the high level of intraoral carbohydrate that is produced by oral intake of these substances. This is in agreement with the observation of Afonsky (17) that salivary carbohydrates, because of the small amount present, probably play an insignificant role in acid formation in the saliva or in the plaque.

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<p>USAF School of Aerospace Medicine, Brooks AF Base, Tex.</p> <p>SAM-TDR-62-140. PAROTID FLUID AND SERUM GLUCOSE LEVELS AND PAROTID FLUID FLOW RATE AS RELATED TO DENTAL CARIES STATUS. Jan. 63, 5 pp. incl. illus., 17 refs.</p> <p>Unclassified Report</p> <p>Parotid fluid samples were collected without exogenous stimulation from 507 systemically healthy young adult males. Blood was drawn from each participant and subjects were classified as to dental caries experience (DMFS). Parotid fluid flow rate</p>	<p>1. Dental caries</p> <p>2. Glucose in parotid fluid</p> <p>I. AFSC Task 775602</p> <p>II. Shannon, I. L., Gibson, W. A., O'Leary, T. J., Chauncey, H. H.</p> <p>III. In ASTIA collection</p>	<p>USAF School of Aerospace Medicine, Brooks AF Base, Tex.</p> <p>SAM-TDR-62-140. PAROTID FLUID AND SERUM GLUCOSE LEVELS AND PAROTID FLUID FLOW RATE AS RELATED TO DENTAL CARIES STATUS. Jan. 63, 5 pp. incl. illus., 17 refs.</p> <p>Unclassified Report</p> <p>Parotid fluid samples were collected without exogenous stimulation from 507 systemically healthy young adult males. Blood was drawn from each participant and subjects were classified as to dental caries experience (DMFS). Parotid fluid flow rate</p>	<p>1. Dental caries</p> <p>2. Glucose in parotid fluid</p> <p>I. AFSC Task 775602</p> <p>II. Shannon, I. L., Gibson, W. A., O'Leary, T. J., Chauncey, H. H.</p> <p>III. In ASTIA collection</p>
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